# Refine Search

**Search Results -**

| Terms | Documents |
|---|---|
| (two adj1 step) adj5 remov$ adj5 solvent | 19 |

**Database:**
US Pre-Grant Publication Full-Text Database
US Patents Full-Text Database
US OCR Full-Text Database
EPO Abstracts Database
JPO Abstracts Database
Derwent World Patents Index
IBM Technical Disclosure Bulletins

**Search:** L4

Refine Search

Recall Text | Clear | Interrupt

## Search History

**DATE: Monday, October 03, 2005** Printable Copy Create Case

| Set Name side by side | Query | Hit Count | Set Name result set |
|---|---|---|---|
| *DB=USPT,EPAB,JPAB,DWPI,TDBD; PLUR=YES; OP=OR* | | | |
| L4 | (two adj1 step) adj5 remov$ adj5 solvent | 19 | L4 |
| L3 | two$step adj5 remov$ adj5 solvent | 6 | L3 |
| L2 | L1 and liposome | 40 | L2 |
| L1 | $step adj5 remov$ adj5 solvent | 2414 | L1 |

END OF SEARCH HISTORY

L2: Entry 15 of 40 File: USPT Apr 10, 2001

DOCUMENT-IDENTIFIER: US 6214300 B1
TITLE: Microencapsulation and electrostatic processing device

Brief Summary Text (10):
Other solid-matrix approaches have utilized copolymers such as polyvinyl chloride/acrylonitrile dissolved initially in organic solvents to form microparticles containing aqueous enzyme solutions. U.S. Pat. No. 3,639,306 to Sternberg et al. discloses a method of making anisotropic polymer particles having a sponge-like inner support structure comprising large and small void spaces and an outer, microporous polymer film barrier. A multiple-step batch process is used which entails removal of the organic solvents used to dissolve the polymers prior to addition of aqueous components.

Brief Summary Text (14):
Certain current methods of forming microcapsules (such as liposomes) are based on chemical characteristics of certain phospholipids that self-assemble into bilayers when dispersed in an excess of water. Most liposomes carry pharmaceuticals dissolved in the entrapped water. Drugs that are insoluble or that have only limited solubility in aqueous solvents pose problems for incorporation into liposomes. Such organic-soluble drugs are usually limited in liposomal formulations to those that bind inside the hydrophobic region of the liposome bilayer. Some drugs are so insoluble that they do not associate with the bilayer and, therefore, have very low encapsulation efficiencies. Certain liposomal drug formulations, including anti-tumor liposomes containing dexorubicin [Gabizion et al. 1992] or muramyltripeptide have been studied extensively in clinical trials. Many conventional therapeutic liposome microcapsules have natural phospholipid outer skins (usually in combination with cholesterol and fatty amine) and therefore are subject to elimination by immune cells. Other conventional methods use sialic acid and other coatings on the lipid bilayer to mask the liposomes from detection by the scavenging immune cells in the reticuloendothelial system (RES).

Brief Summary Text (15):
Conventional methods of forming microcapsules are based on liquid--liquid dispersions of aqueous drugs and organic solvents. The dispersion methods often require emulsification of the aqueous phase into organic carrier solutions by shear, bubbling or sonication. These methods typically produce water-in-oil (W/O) type liposomes, for which a second requisite step is the removal of the organic solvent (typically by evaporation) to form reverse-phase evaporation vesicles or stable plurilamellar vesicles. The size distribution for these vesicles is quite heterogeneous.

Brief Summary Text (17):
For instance, U.S. Pat. No. 4,855,090 to Wallach, discloses a method of making a multilamellar lipid vesicle by blending an aqueous phase and a nonaqueous lipophilic phase using a high shear producing apparatus. The lipophilic phase is maintained at a high temperature (above the melting point of the lipid components) and is combined with an excess of the aqueous phase, which is also maintained at a high temperature. U.S. Pat. No. 5,032,457 to Wallach discloses a paucilamellar lipid vesicle and method of making paucilamellar lipid vesicles (PLV). The method comprises combining a nonaqueous lipophilic phase with an aqueous phase at high

temperatures and high shear mixing conditions, wherein the PLVs are rapidly formed in a single step process. U.S. Pat. No. 4,501,728 to Geho et al. discloses the encapsulation of one or more drugs or other substances within a liposome covered with a sialic acid residue for masking the surface of the membrane from scavenging cells of the body utilizing techniques known for the production of liposomes. In one embodiment, additional tissue specific constituents are added to the surface of the liposome which cause the liposome thusly treated to be attracted to specific tissues. Similarly, U.S. Pat. No. 5,013,556 to Woodle et al. provided methods for making liposomes with enhanced circulation times. Liposomes created by this method contain 1-20 mole % of an amphipathic lipid derivatized with a polyalkylether (such as phosphatidyl ethanolamine derivatized with polyethyleneglycol). U.S. Pat. No. 5,225,212 to Martin et al. discloses a liposome composition for extended release of a therapeutic compound into the bloodstream, the liposomes being composed of vesicle-forming lipids derivatized with a hydrophilic polymer, wherein the liposome composition is used for extending the period of release of a therapeutic compound such as a polypeptide, injected within the body. Formulations of "stealth" liposomes have been made with lipids that are less detectable by immune cells in an attempt to avoid phagocytosis [Allen et al. 1992]. Still other modifications of lipids (i.e., neutral glycolipids) may be affected in order to produce anti-viral formulations (U.S. Pat. No. 5,192,551 to Willoughby et al. 1993). However, new types of microcapsules are needed to exploit the various unique applications of this type of drug delivery.

Brief Summary Text (21):
Allen, T. M., Mehra, T., Hansen, C. and Chin, Y. C., Stealth Liposomes: An Improved Sustained Release System for 1-b-D-Arabinofuranosylcytosine, Cancer Res. 52:2431-39, 1992.

Brief Summary Text (22):
Gabizon, A., et al., Liposome-Associated Doxorubicin: Preclinical Pharmacology and Exploratory Clinical Phase, in G. Lopez-Berestein and I. J. Fidler (Eds.) Therapy of Infectious Diseases and Cancer, Alan R. Liss, Inc., New York, pp. 189-203, 1992.

Brief Summary Text (23):
Talsma, H. and Crommelin, D. J. A., Liposomes as Drug Delivery Systems, Part 1: Preparation. Pharmaceutical Technology, pp. 96-106, October 1992.

Previous Doc Next Doc Go to Doc#

L2: Entry 29 of 40 File: USPT Apr 18, 1995

DOCUMENT-IDENTIFIER: US 5407660 A
TITLE: Diagnostic liposomal compositions for enhancing NMR imaging

Abstract Text (1):
Nuclear magnetic resonance (NMR) imaging of body organs and tissues is enhanced by administering to a living animal body a diagnostic composition comprising a diagnostically effective amount of a substantially nontoxic paramagnetic image altering agent containing a chelate of a paramagnetic element such as magnesium, gadolinium or iron, carried by a liposome. The chelate is carried by or within or outside the external surface of the liposome in such a manner that after arrival at or delivery to the desired organ or tissue site, the paramagnetic image altering agent is released in a diagnostically useful fashion.

Brief Summary Text (14):
Briefly, the invention is directed to a method for enhancing NMR imaging of body organs and tissues which comprises administering a substantially nontoxic paramagnetic image altering agent to a living animal body in a sufficient amount to provide enhancement of NMR images of said body organs and tissues, the substantially nontoxic paramagnetic image altering agent containing a chelate of a paramagnetic element carried by a liposome.

Brief Summary Text (20):
Further, in order to improve the organ target specificity of such chelates while retaining the advantageous low toxicity thereof, it has been found desirable in accordance with the present invention to administer the paramagnetic image altering agent in a form in which the chelate of a paramagnetic element is carried to a desired site by means of a liposome. Such preparations are particularly suitable for enhancement of NMR images of the reticuloendothelial system (RES) since the liposome provides greater liver/spleen specificity by permitting the chelate to be present in such organs in greater concentrations and for longer residence periods than would otherwise be found with the chelate alone. The liposome functions as a carrier for delivering the paramagnetic image altering chelate to the desired organs without itself significantly altering the NMR proton signals. A typical preparation of this nature is disodium (ethylenediaminetetraacetato) manganese (II) contained in solution and within multilamellar liposomes as illustrated in greater detail hereinafter. The chelate is carried by or within or outside the external surface of the liposome in such a manner that after arrival at or delivery to the desired organ or tissue site, the paramagnetic image altering agent is released in a diagnostically useful fashion.

Brief Summary Text (21):
Liposomes generally comprise lipid materials including lecithin and sterols and the liposomes employed herein may contain egg phosphatidyl choline, egg phosphatidic acid, cholesterol and alpha-tocopherol in various molar ratios and the lipids may be present at various total concentrations. Useful liposomes may be prepared as generally described in Kimelberg et al., CRC Crit. Rev. Toxicol. 6 25 (1978), Papahadjopoulos, Ann. Reports in Med. Chem., 14 250-260 (1979) and Olson et al., Biochim. Biophys. Acta., 557 9-23 (1979). The preparation of liposomes and their release and stability characterics are also described in Yatvin et al., Medical Physics, Vol. 9, No. 2, 149 (1982).

Detailed Description Text (10):
A liposome preparation of disodium (ethylenediaminetetraacetato)manganese (II) (EDTA) (composition A in Example 1) for use in NMR imaging was prepared according to the following procedure.

Detailed Description Text (11):
Egg phosphatidyl choline (396 mg) (PC) (type V-E, Sigma Chemical Co.), dipalmitoyl phosphatidic acid (85.6 mg) (PA) (Sigma) , cholesterol (153.4 mg) (CH) (Sigma) , and alphatocopherol (14.14 mg) (a-T) (Sigma) were combined in a 150 ml glass round-bottom flask. This was done by dispensing appropriate volumes of stock solutions of these compounds (chloroform-methanol solutions stored at -15.degree. C.). Total solvent volume in the flask was 75.8 ml at this point. An additional 50 ml chloroform was added, and then the flask was placed on a rotary evaporating unit (Buchi, Type KRvr) utilizing dry ice-acetone in the condenser. A water bath (approx. 45.degree. C.) was placed under the flask, in contact with the lower third of the flask. The solvents were then dried off as the evaporator vacuum was raised gradually. The lipids were dried to a thin, even film covering approximately one half of the flask area. This process required approximately 15 minutes. The flask was then removed from the evaporator and connected directly to the laboratory vacuum. The purpose of this step was to ensure removal of residual organic solvents. This vacuum step was done for approximately 1 hour at room temperature. For this preparation, the flask was then taken off the vacuum, stoppered, then placed at -15.degree. C. overnight. On the following day, the flask was placed on the laboratory vacuum at room temperature for 1 hour. Then 55 ml of composition A of Example 1 was dispensed into the flask and the flask was stoppered. The flask was then swirled by hand so that the solution was swept across the dried lipid. The lipid gradually became suspended in the solution, and the end point was that at which all the lipid had been visually dispersed from the wall of the flask. This required 45 minutes for this preparation. At this point, the liposomes had been formed (Kimelberg et al., "Properties and Biological Effects of Liposomes and their Uses in Pharmacology and Toxicology", CRC Crit. Rev. Toxicol. 6 25 (1978) and Papahadjopoulos, "Liposomes as Drug Carriers", Ann. Reports in Med. Chem., 14 250-260 (1979)). For this preparation, the liposome lipid composition was egg PC/PA/CH/a-T=8/2/6/0.5, expressed on a molar ratio basis, and the total lipid concentration was 20 micromoles/ml. The liposomes were then transferred to a glass beaker and then aspirated into a 25 ml glass syringe with a luer fitting. A 25 mm Swinnex filter housing (Millipore Corp.) was then connected to the syringe. The housing had previously been fitted with a 25 mm 1.0 micron pore size Unipore membrane with a polyester post filter (both from Bio-Rad Corp.). The liposomes were then extruded through the membrane by depressing the syringe plunger. This process was repeated until all 55 ml were extruded. This step was done to narrow the liposome size distribution (Olson et al., "Preparation of Liposomes of Defined Size Distribution by Extrusion through Polycarbonate Membranes", Biochem. Biophys. Acta., 557 9-23 (1979)). There is no retention of lipid by the membrane and so the liposome lipid concentration remains the same. The sizing effect is presumably done by breaking the larger liposomes down to smaller ones as they pass through the membrane pores. Fifty ml of the liposomes were then placed in a 50 ml glass vial with an 890 gray stopper. The vial was placed in a refrigerated container for use in NMR imaging.

Detailed Description Text (13):
A liposome preparation of manganous disodium ethylenediaminetetraacetate (composition B in Example 1) for use in NMR imaging was prepared according to the following procedure.

Detailed Description Text (14):
Egg phosphatidylcholine (769 mg), egg phosphatidic acid (172 mg) (Avanti Polar Lipids, Inc.), cholesterol (446.5 mg), and alpha-tocopherol (27.55 mg) were combined in a 250 ml round-bottom flask. Solvent volume at this point was 179 ml.

The solvents were then dried, and the lipids deposited on the flask walls, in the same manner as for the Example 2 batch. The dried lipids were placed on the laboratory vacuum as before, and then immediately afterward, 48 ml of composition B of Example 1 were added to the flask, and the lipid was dispersed as described previously in Example 2. The dispersal process required 1.5 hours to complete, and 10-15 small glass beads (2 mm diameter) were used during the last 5 minutes to help disperse the lipid. For this preparation, the liposome lipid composition was egg PC/egg PA/CH/a-T=8/2/9/0.5, and the lipid concentration was 50 micromoles/ml. The liposomes were then extruded, in the same manner as before, through a 1.0 micron pore size Unipore membrane. Thirty minutes prior to this, several 6-inch lengths of dialysis tubing (Spectrapor, 1 inch width, 10,000 MW, Spectrum Medical Industries, Inc.) were placed in 0.9% NaCl to hydrate. After extrusion, the liposomes were transferred to these bags (5 required) and the bags were clamped off. Approximately 0.5 ml of liposomes were held aside. The bags were placed in a 4 L beaker containing 3.8 L of 0.9% NaCl, pH 6.4. The bags float because of the plastic dialysis bag clamps used. A magnetic stirring bar was placed in the beaker, and the beaker was covered with aluminum foil and placed on a magnetic stirrer (Thermodyne Inc., Type Nuova II) in a 5.degree. C. cold room. The stirrer was turned up to the point at which the bags were gently agitated. After 18 hours, the bags were placed in a beaker of fresh, precooled 0.9% NaCl, and the dialysis was continued. After 23.5 hours, the beaker was removed from the cold room, the bags were cut open, and the liposomes were transferred to a previously cooled 50 ml glass vial and stoppered. The vial was placed in a refrigerated container for use in NMR imaging. The purpose of this dialysis step was to remove the non-liposome-entrapped Mn-EDTA from the preparation. The Mn-EDTA concentration in the entrapped aqueous solution remained the same as it was before dialysis, but the external (i.e. non-liposome-entrapped) aqueous solution now consisted of just 0.9% NaCl.

Detailed Description Text (16):
Acute intravenous toxicity testing was performed in mice with manganese (II) chloride (MnCl.sub.2), composition B of Example 1 (containing 4.6% manganous disodium ethylenediaminetetraacetate with 0.67% calcium disodium ethylenediaminetetraacetate), the composition of Example 2 and a concentrated aqueous suspension of the negatively charged liposomes alone in phosphate-buffered saline, test substances 1 through 4, respectively. In addition, the cardiotoxic effects of single bolus intracoronary arterial injections of 4, 8, 16 or 32 mg MnCl.sub.2 were evaluated in the isolated perfused rabbit heart (IPRH).

Detailed Description Text (17):
The manganese (II) chloride was dissolved in sterile water for injection, USP (SWFI; Abbott Laboratories) to yield unhydrated salt concentrations of 0.25%, w/v and 0.8% w/v for the mouse and IPRH testing, respectively. The aqueous vehicle for the suspension of negatively charged liposomes consisted of 0.9% NaCl buffered with 0.003 M sodium phosphate, pH 7.4. The lipid concentration was 50 .mu.mol/ml.

Detailed Description Text (29):
Neither test substance 2 (MnNa.sub.2 EDTA) nor test substance 3 (MnNa.sub.2 EDTA/liposome formulation) caused death at doses up to 2300 mg/kg of MnNa.sub.2 EDTA. Larger doses were not given because dose volumes would have been excessive. However, the 2300 mg/kg dose of MnNa.sub.2 EDTA did appear to impair weight gain as evidenced by data for both test substances 2 and 3. There were also slight increases or decreases in motor activity in a majority of mice receiving test substances 2 and 3 at all dose levels. The 2300 mg/kg dose, which is obviously considerably lower than the LD.sub.50, corresponds to 6.03 mmol/kg of Mn(II).

Detailed Description Text (30):
Disodium (ethylenediaminetetraacetato) manganese (II) (test substance 2) had a much lower degree of acute intravenous toxicity than MnCl.sub.2. There were no deaths at a dose of test substance 2 which was 27 times as great as the LD.sub.50 dose of MnCl.sub.2 when compared on the basis of manganous content. The acute toxicity of

disodium (ethylenediaminetetraacetato)manganese (II) did not appear to be changed by partial incorporation (5%, v/v, entrapped) into negatively charged liposomes (test substance 3). A concentrated suspension of liposomes alone failed to elicit any toxic effects following intravenous administration to mice (test substance 4).

Detailed Description Text (31):
Test substance 4, a phosphate buffered saline suspension of liposomes with 50 .mu.mol lipid/ml, was nontoxic in doses up to 50 ml/kg, a dose considered to be the maximal safe dose volume. No adverse reactions were observed and body weight gain was normal. Test substance 4 contained 2.5 times the lipid content of test substance 3 in an attempt to increase the likelihood that liposome-induced toxicity might be manifested.

Detailed Description Text (38):
A study was carried out to compare the biodistribution of disodium (ethylenediaminetetraacetato) manganese (II) (.sup.54 MnNa.sub.2 EDTA) and a partially liposome-entrapped manganous disodium (ethylenediaminetetraacetato) manganese (II) formulation (.sup.54 Mn-L).

Detailed Description Text (62):
3. A negatively charged liposome formulation containing encapsulated .sup.54 MnNa.sub.2 EDTA. The liposomes consisted of phosphatidylcholine, phosphatidic acid, cholesterol and .alpha.-tocopherol (8/2/9/0.5) and were dispersed in Test substance 1 (12.5 .mu.Ci/ml). Liposomes were dialyzed against normal saline prior to injection in order to remove unencapsulated .sup.54 MnNa.sub.2 EDTA. Greater than 98.5% of the radioactivity was entrapped following dialysis. The liposome entrapment of Test substance 1 was 9.0%, v/v, and the lipid concentration was approximately 50 .mu.mol/ml.

Detailed Description Text (63):
4. A positively charged liposome formulation containing encapsulated .sup.54 MnNa.sub.2 EDTA. The liposomes consisted of phosphatidyl choline, stearylamine, cholesterol and .alpha.-tocopherol (8/2/9/0.5) and were dispersed in Test substance 1 (12.5 .mu.Ci/ml). Liposomes were dialyzed against normal saline prior to injection in order to remove unencapsulated .sup.54 MnNa.sub.2 EDTA. Greater than 98.5% of the remaining radioactivity was entrapped following dialysis. The liposome entrapment of Test substance 1 was 12.8%, v/v, and lipid concentration was approximately 50 .mu.mol/ml.

Detailed Description Text (65):
The rats of each of four treatment groups (each group consisting of 10 males and 10 females) received single 2 ml/kg intravenous injections of each of the four test substances. Animals were sacrificed at 0.5, 2, 4, 24 and 48 hours after treatment and an extensive number of tissues were sampled for radioactivity determination. In addition, urine samples were obtained from rats sacrificed at time intervals >0.5 hour and fecal samples at 24 and 48 hours after treatment. Urine samples collected from rats treated with test substances 1 and 2, sacrificed at 48 hours were analyzed by paper chromatography for the presence of radioactive metabolites. The unencapsulated solutions (test substances 1 and 2) accumulated on a dose-dependent, apparently saturable, basis primarily in liver, small intestine, bone and marrow, muscle and kidney. The liposome entrapped solutions (test substances 3 and 4) accumulated in extremely high concentrations in the liver and spleen indicating uptake by the reticuloendothelial system. In addition, relatively high concentrations of all four formulations tested were observed in glandular organs such as the pancreas and salivary gland. The unencapsulated formulations (test substances 1 and 2) tended to undergo faster tissue elimination than the entrapped formulations (test substances 3 and 4).

Detailed Description Text (66):
Test substances 1 and 2 were eliminated from the liver, kidney and spleen via

biphasic kinetics indicating heterogeneous cellular compartmentalization of the .sup.54 MnNa.sub.2 EDTA. Test substances 3 and 4 were eliminated from most organs slowly via monophasic kinetics indicating homogenous cellular distribution. In addition, the negatively charged liposome formulation (test substance 3) was more rapidly cleared from the liver and radioactivity accumulated faster in the small intestine compared to test substance 4.

Detailed Description Text (77):
The preparation of Example 2 with manganous disodium ethylenediaminetetraacetate contained in solution within and outside multilamellar liposomes was administered to an anesthetized dog. The total dose was 25 ml of the preparation, 12 ml of which was administered by bolus injection and the remaining 13 ml by infusion over a 3-5 minute period. The dog was positioned on his back in a General Electric NMR clinical scanner. The desired level for transverse scanning was determined prior to the administration of the NMR enhancing preparation.

Detailed Description Text (94):
A rabbit received 10 ml/kg (approximately 0.1 mmol Mn(II)/kg) of a preparation with disodium (ethylenediaminetetraacetato)manganese (II) contained in solution within multilamellar liposomes (MnNa.sub.2 EDTA liposomes, test substance 3 of example 6). The rabbit was then anesthetized, placed in the General Electric NMR scanner and images of the abdominal area, including liver and spleen, were obtained over a period of 2-5 hours after dosing. Liver and spleen images were altered as compared to similar scans in untreated rabbits, while no effect was observed in intestine, kidney and skeletal muscle. The peak alterations of liver and spleen proton signals were observed at the beginning of the imaging period (two hours after dosing with the liposomal formulation).

Detailed Description Text (96):
Groups of two rabbits each received intravenous doses of 3 ml/kg or 10 ml/kg (approximately 0.03 or 0.1 mmol Mn(II)/kg) of a preparation with disodium (ethylenediaminetetraacetato)manganese (II) contained in solution within multilamellar liposomes (MnNa.sub.2 EDTA liposomes, test substance 3 of example 6) and were sacrificed at intervals of 2-14 hours after dosing. Liver and spleen proton T.sub.1 values were determined using a 0.12 T NMR spectrometer with a probe frequency of 5.1 mHz. Dose-related decreases in liver and spleen T.sub.1 values were observed with peak effects at two hours (see following table):

Detailed Description Text (98):
The plasma fragility of test substances 3 and 4 of example 6 was tested as follows: 0.5 ml each of liposomes and fresh dog plasma were mixed, then placed at 37.degree. C. for 30 minutes. The mixture was then cooled and an aliquot was then run on a Sephadex G-50 column to separate the unentrapped (released) material from the liposome entrapped material. In both cases approximately 10-15% leakage had occurred, indicating that the liposomes were stable enough to use for in vivo imaging.

Detailed Description Paragraph Table (8):
MEAN TISSUE T.sub.1 VALUES (msec) AT VARIOUS TIMES AFTER NORMAL ADMINISTRATION OF TISSUE MnNa.sub.2 EDTA LIPOSOMES DOSE TISSUE T.sub.1 (msec) 2 hrs 4 hrs 12-14 hrs 3 ml/kg spleen 260 99 133 -- liver 171 99 131 -- 10 ml/kg spleen 260 35 45 80 liver 171 93 110 120

CLAIMS:

1. A diagnostic composition for enhancing NMR imaging of body organs and tissues comprising a diagnostically effective amount of a substantially nontoxic paramagnetic image altering agent containing a chelate of a paramagnetic element, said paramagnetic image altering agent being carried by or within or outside the

external surface of a liposome.

Previous Doc Next Doc Go to Doc#